

# BIOLOGICAL CLOCKS

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FRONT COVER      A fiddler crab. This crab shows an interesting rhythm in body colour changes. At dawn it starts to grow dark in colour. After sunset it rapidly becomes lighter in colour.

BACK COVER      The sleep movements of the leaflets of the common Indian rain tree. The leaflets close and droop regularly at dusk giving the impression that the tree has gone to sleep.

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## Foreword

THE usefulness of science as a subject of study has indeed been recognised during the last few decades. But now, more than ever, when the country has launched tremendous economic and social welfare programmes which hinge upon the application of the latest advances in science and technology, it has become essential that every citizen should have as much knowledge of science as possible. While science is now being made a compulsory subject at the school level and textbooks based on new concepts of science teaching are being introduced, a textbook can only impart knowledge within the limits of the school syllabus. Therefore, in an attempt to make easily accessible to young minds additional and more comprehensive information in the various areas of science, the National Council is preparing and bringing out a series of supplementary reading materials on science. The present book is one of this series, and I hope that it will be found useful not only by students but by teachers and others as well.

RAIS AHMED

*Director*

New Delhi

18 February 1975

National Council of Educational  
Research and Training



## Preface

A CLOCK has always meant a device to indicate the passage of time. It has, however, been seldom realized that all organisms have also some kind of a built-in mechanism to measure time. In their turn, organisms are also subjected to several environmental changes occurring sequentially like light and darkness, or high and low temperatures. It is almost trite to be reminded of the crowing of the cock heralding dawn or the display of the brilliant plumage of the peacock at the sight of rain-bearing clouds, to indicate such mechanisms.

The adaptability of organisms to their environment *vis-a-vis* the regulation of their built-in physiological clock work is a field of interesting study. From a biological view-point the survival of an organism is the true test for the success of the physiological adaptation of an organism in relation to environmental changes. The regularity of the occurrence of these adjustments constitutes a "biological rhythm". The nature, duration, pattern and range of these rhythms are indeed variable; so much so that a comprehensive school of biology has now sprung up the world over, to study these phenomena. Such rhythms in animals appear to be more widespread because these studies are correlated to an understanding of the nervous and chemical coordination mechanisms present in animals. In plants, however, the mechanisms underlying the rhythms appear to have more of a molecular and  $\therefore$  chemical base.

The famous book of H. G. Wells, *The Time Machine*, and the works of Aldous Huxley such as *Time must have a stop*, *Ape and Essence* and *Brave New World* need perhaps be no longer considered satirical reflections on science; on the other hand, a study of biological rhythms in this background has far more serious, yet definite, implications underlying the very basic philosophy of science

Dr R. Narayanaswamy is one of our younger generation of scientists endowed with a deep comprehension of the basic phenomena of the life processes and a clear discernment of vital biological principles like the biological rhythms. It is, indeed, fortunate that we have once again an author of such accomplishments to contribute a title to our series of background reading booklets in biology

The brief, yet incisive, account of the "Biological Clocks" is one more attempt to enthuse the young children and the lay citizens of our country to take a greater interest in the many facets of biological studies, so that the scientific edifice of this country will rest on firm and certain foundations

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## Introduction

MAN has devised clocks and calendars, which enable him to determine the time of the day or the seasons. It may not be widely known that living things at all levels of organization have built-in mechanisms to measure out the passage of time. Organisms have always been subjected to the daily and seasonal changes of light and darkness and of high and low temperatures. Organisms are remarkably adapted to these environmental changes and show within themselves, in their physiological processes, many rhythmic patterns. Some of these are easily observed. For instance, everybody is familiar with the regular opening and closing of certain flowers at particular times of the day as well as the upward and downward movements of the leaves of some plants of the bean family (*see*

cover). If such leaf movements are plotted in the form of a graph with time shown along the

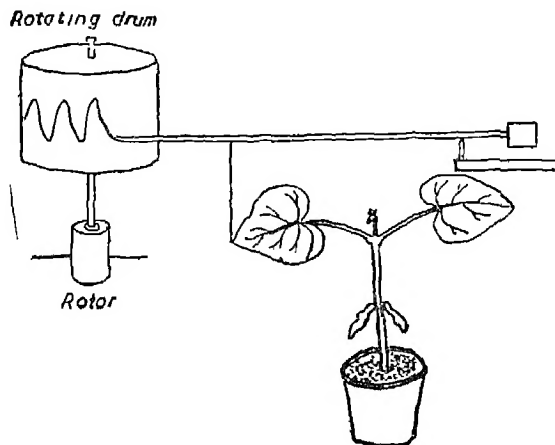
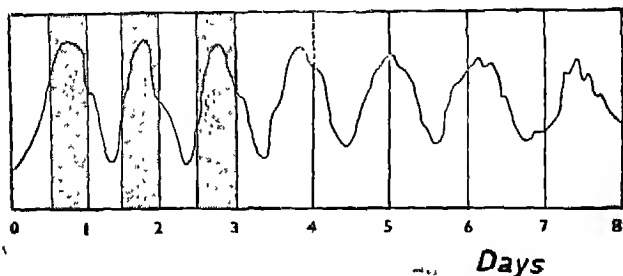


Fig 1. Device to record leaf movements. The clock work in the rotor rotates the drum at a slow and uniform rate. A sheet of paper is wound round the drum on which a pen records the movements of the leaf. The leaf is attached to the pen through a lever mechanism. The whole set-up can be placed under controlled conditions.

Adapted from A. EMME, *The Clock of Living Nature*, Peace Publishers, Moscow, p. 54

X-axis and the position of the leaf along the Y-axis, it will be observed that the peaks and

troughs in the curve will occur at regular intervals (*Fig. 2*). These rhythms occur not only



**Fig 2.** Sleep movements of a beech leaf shown graphically (recorded by a device of the type shown in *Fig 1*) During the first three days alternating light and dark periods were provided and afterwards the plant was in continuous light. Note that the rhythm continues under continuous light and that the peaks occur at regular intervals

Adapted from W A JENSEN & L. G KAVALJIAN (ed), *Plant Biology Today*, Macmillan & Co., Ltd, London, fig. 4-2

Original E BÜNNING, 'Mechanismus und Leistungen der physiologischen Uhr', *Nova Acta Leopoldina*, N. F Vol 21 (1959), p 181

under natural conditions where night and day alternate but also under constant conditions of continuous light or continuous darkness, at least for some time. Hence these rhythms are referred

to as *endogenous* to distinguish them from rhythms that may be induced or maintained by alternation of night and day. Under constant conditions, these rhythms have periods varying somewhat from 24 hours and are therefore referred to also as *circadian* (*circa* about, *dies* day) rhythms.

The endogenous diurnal rhythms were first observed in plants and the earliest record of the leaf movements in plants of the bean family goes back to the days of Alexander, the Great ! In the eighteenth century, these leaf movements were again studied and an astronomer, De Mairan by name, found that the movements of the leaves continued even in constant darkness, the first demonstration that the rhythm was endogenous. Investigations of endogenous diurnal rhythms in animals began later. Some examples are given in the next chapter.

A very interesting and important feature of these rhythms is that the period of each rhythm is remarkably constant. The status of the rhythm at any particular moment (for example, the position of the leaf blade) might serve to determine the time of the day and the uniform change of the rhythm to meter out the passage

of time. Hence such a rhythm resembles a clock mechanism and has been called a biological or physiological clock.

It was mentioned earlier that under constant conditions the period of physiological rhythms shows deviations from the 24-hour period. It could be anywhere between 22 and 28 hours. The length of the period is, however, characteristic of the particular individual being studied and remains strikingly constant. It has been shown that this character is inherited. Maintaining the organisms under constant conditions from the earliest stages of development or even through several generations does not alter the period of the rhythm.

## 2

### **Diurnal Rhythms in Different Organisms**

CIRCADIAN RHYTHMS have been detected in living beings at all levels of organization. As you might be aware the simplest of living beings are made up of a single cell. At a slightly higher level are the multicellular plants like many algae, fungi and bryophytes, followed by ferns and higher plants. Among animals also we have a whole range from unicellular forms to the most advanced mammals like the primates. Even the simplest of plants and animals show rhythmic behaviour. Although information on bacteria on this aspect is limited, in one case at least it has been shown that growth rate fluctuates diurnally under constant conditions. There are in the oceans of the world large floating masses of tiny plants and animals which consti-

tute the main source of food for the fishes in the seas. These floating organisms are known as plankton (phytoplankton or zooplankton). These planktonic organisms, some of which are unicellular and others multicellular, exhibit regular vertical migration every 24 hours. At dusk they migrate upwards and become concentrated in the surface layer. Then at midnight they sink to a considerable depth. Again at dawn they rise once more to a level where the light intensity (which constitutes the stimulus for these migrations) is optimum. When the light intensity increases as the sun moves across the heavens, there is again a descent to a lower level. In the unicellular *Euglena*, an organism extensively used in physiological experiments, a pronounced variation in the ability to move in response to the light stimulus has been observed even in continuous darkness of very long duration. Another well-known unicellular alga which shows various kinds of diurnal rhythms and which has been studied extensively is *Gonyaulax polyedra*. This alga is what is known as a dino-flagellate, has two flagella or whip-like organs and a cell wall made up of a number of plates of cellulose fitted together to form a mosaic (Fig. 3). This organism is common

in the oceans and has the ability to give out flashes of light like the firefly Enormous

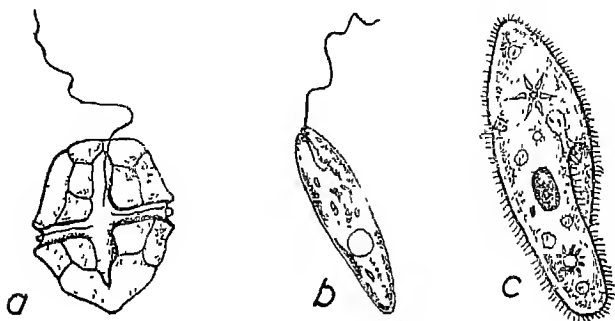


Fig. 3. Some unicellular organisms showing circadian rhythms. (a) *Gonyaulax polyedra* (b) *Euglena* (c) *Paramecium*

Adapted from: N J BERRIL, *Biology in Action*, Heinemann Educational Books Ltd., London, figs 11-2 and 11-5

numbers of this organism, together with others, cause a beautiful glow at nights in the ocean in certain parts of the world. Studies by scientists in the United States have shown that there is a definite diurnal rhythm in the ability of this alga to emit flashes of light (known as bioluminescence). Luminescence occurs essentially at night and continues with the same periodicity even when the organism is grown continuously



in light of low intensity. There is also a regular diurnal rhythm in photosynthesis (the peak occurring during day-time) and in cell division in this alga (*Fig. 4*). In *Oedogonium*, a filamentous alga, there is a rhythm in the discharge of spores, i.e. special cells which grow into new individuals. This takes place every 22 hours. A unicellular animal commonly found in water is *Paramecium*, also known as 'slipper animalcule' because of its shape. During sexual reproduction in this organism two individuals come together and fuse in a process known as conjugation. There is a definite diurnal rhythm in this process.

There is a large group of colourless plants commonly called fungi. Among members of this group also rhythmical behaviour has been observed. Many fungi reproduce by producing small bodies called spores. In some cases, these spores are released in greater abundance at a particular time during the 24-hour period (say, at midnight) and this goes on even in continuous darkness.

Circadian rhythms in higher plants are more common and more easily observed. The rhythmic movement of the leaf blades in

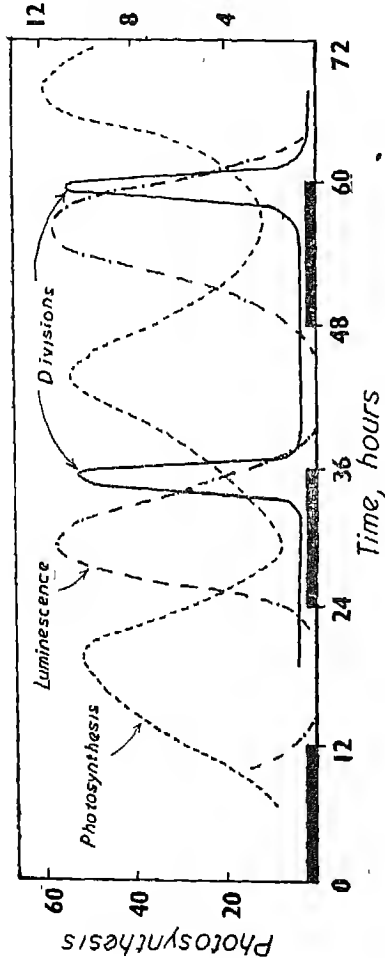
*Luminescence or divisions*

Fig 4. Diurnal rhythms of luminescence, photosynthesis and cell divisions in cultures of *Gonyaulax polyedra* kept under conditions of alternating light and dark periods. The shaded rectangles along the X-axis represent dark periods. The rhythms continue even under constant conditions

Adapted from J. W. HASTINGS, "Unicellular Clocks",  
*Annu Rev Microbiol.* 13, 1959, fig 1, Annual Reviews  
 Inc, Palo Alto, Calif, USA

plants of the bean family has been referred to earlier. Most of you would have observed the sleep movements of the leaflets of the common Indian rain tree (*shown on the cover*). Here the leaflets close and droop regularly at dusk giving the impression of the tree having gone to sleep. Petals of many kinds of flowers open and close with a diurnal rhythm. Some flowers like the night queen have rhythms in odour production, and consequently one suddenly feels its fragrance in the air at a particular time after dusk every day. Flowers show rhythmic behaviour in nectar secretion also. If the stem of a sunflower plant is cut, sap exudes from the cut stump. If such a stump is placed in constant darkness and temperature and the rate of exudation measured (as volume of sap exuded per hour) it will be seen that the rate is not uniform but increases to a maximum during the period corresponding to midday and decreases to a minimum coinciding with midnight. This goes on day after day.

Examples of diurnal rhythms in animals are varied and numerous. It is common knowledge that some animals are nocturnal in habit while others are active during day-time. This

is a typical and simple example of rhythmic behaviour. For instance, household pests like cockroaches and rats are seen to come out of their hiding-places and to be active only at night. These organisms have been kept under constant conditions of temperature and light with special devices to record their movements and activity continuously. Such experiments have revealed that their activity is pronounced only during a part of the 24-hour period and that such spurts of activity recur at regular intervals. Bats, owls, snakes, etc., also exhibit nocturnal activity. The activity rhythms of animals are usually associated with their feeding rhythms. With us, the need for food is sometimes a matter of habit. Even bees, if they are offered food at a fixed time for several days, will continue to search for food at this particular time even if it is no longer available. It would appear as if a timing mechanism has been set in motion inside these insects.

Most people are aware that the crowing of cocks and the cawing of crows herald sunrise. Some innate clock mechanism in these birds seems to enable them to sense that daybreak is close at hand. It is no wonder that for the rural

folk a cock is as good as an alarm clock. Some of you might be familiar with the fruit-fly. Many of the ideas of modern genetics have emerged from work on these insects. The young insects of this species emerge from the pupal stage almost invariably at dawn. In many other insects also this is the case. We shall refer to some other interesting aspects of this phenomenon—the time sense of insects—at a later stage. In experiments with certain beetles, a daily rhythm in susceptibility to some insecticides (poisons used to kill insect pests) has been observed. The beetles were always more resistant at dawn and became very susceptible within three hours after dawn.

There is a rhythm in body colour change in certain crabs like the fiddler crab (*Fig. 5*). At dawn the crab starts to grow dark in colour. This helps it to escape from predatory animals and to protect itself from intense light. After sunset the crab rapidly becomes lighter in colour. These changes in colour are due to migration of pigment into or away from certain cells in the body surface. This phenomenon has been studied extensively and shown to have an endogenous rhythm.

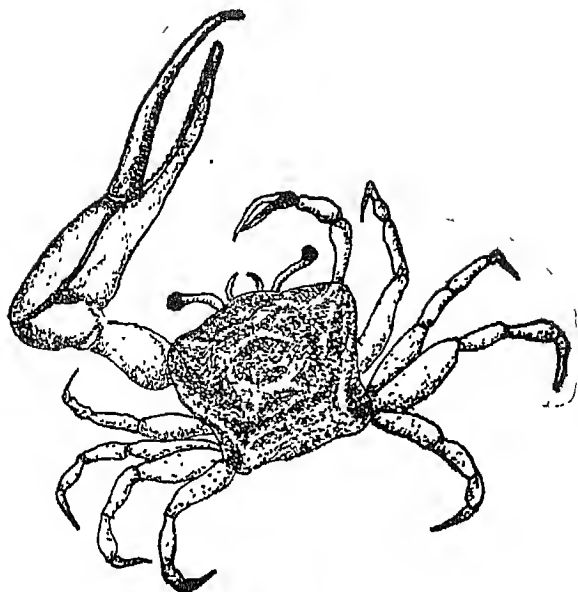


Fig. 5. Fiddler crab. This crab shows an interesting rhythm in body colour changes as described in the text.

Various kinds of rhythmic phenomena can be observed in human beings. A very familiar example of the innate time-sense in some people is their ability to wake up at a fixed time every morning without the aid of an alarm clock. This

is referred to as a 'head clock'. Every one who has been ill at any time would have noticed that the body temperature goes up during evening and comes down in the morning. This is due to a diurnal fluctuation in body temperature which is present, though not noticed, even in healthy

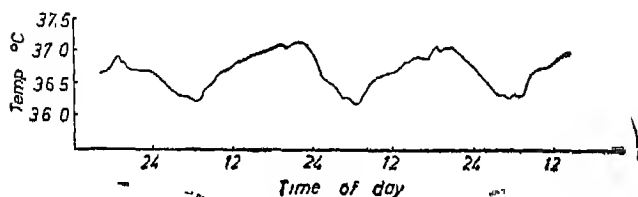



Fig 6 Diurnal fluctuations in body temperature in man

Adapted from E BUNNING, *The Physiological Clock*, Longmans Springer-Verlag, New York, 1967, fig 43

individuals (Fig. 6). Similarly, changes occur in the proportion of the various kinds of corpuscles in the blood in the course of 24 hours. Rhythms of wakefulness and sleep develop quite early in life at about the time when a baby is 6-7 weeks old and persist throughout life

The diurnal changes in metabolic processes in the human being is of great significance in certain diseases. For instance, in persons suffer-

ing from asthma, attacks are generally nocturnal or more severe at night than during daytime. It is well known that asthmatic attacks can be alleviated by giving the patient corticosteroids, a kind of hormone secreted by the adrenal glands. Blood levels of these hormones are low at night and this seems to coincide with asthma attacks. Sugar, chloride and cholesterol contents in blood and blood pressure also show rhythmic changes. Many psychiatric disorders are characterized by rhythmic appearance of symptoms. In some cases of epilepsy, fits occur daily at regular times. In some mental illnesses abnormalities have been seen to appear on alternate days.





## Utility of the Clock

THE VARIOUS KINDS of physiological rhythms could serve as a clock mechanism for the organism concerned to measure out time. The importance of time measurement for any living being cannot be overstressed. It enables plants and animals to perform certain activities at a certain time of the day which is most suitable for such activity. The rhythm in photosynthesis, for instance, enables the most optimum use of the factors favourable for the process. The sleep movements of the leaves, perhaps, enable the plants concerned to cut down water loss. There is no need for the leaves to be held in a position for receiving radiant energy after the sun has set. The emergence of young insects from the pupae at daybreak is definitely advantageous as the freshly emerged insects are

thereby not exposed immediately to the drying effects of sunlight. The time (night) when the filarial parasite is present in large numbers in the blood of the victim coincides with the period of feeding by mosquitoes which transmit the disease. The rhythm of activity of various animals is obviously connected with the availability of food. The bees seem to know the time when the flowers they feed upon will be open and secrete nectar. The rhythm of discharge of the sex cells by certain sea weeds enables a high concentration of both kinds of gametes to be present at the same time, thereby increasing the chances of fertilization.

The practical applications of a knowledge of these rhythmic phenomena will be discussed later.

## Some Features of the Endogenous Rhythms

It was mentioned earlier that when an organism is placed under constant conditions the rhythms of various physiological processes cease to show a strict 24-hour periodicity but have periods of *approximately* 24 hours, the length of the period being characteristic of the organism concerned. Another feature of the behaviour, under constant conditions, is that, in most instances, the rhythm fades out within a few days or weeks. For example, a cockroach placed in continuous darkness will cease to exhibit definite periods of activity alternating with periods of rest. Similarly, if organisms are raised under constant conditions from the earliest stages, they fail to show rhythmic behaviour. In both these cases, i.e. where the rhythms have not developed at

all, as well as where the rhythms have faded out, rhythmic behaviour can be induced or reinitiated, as the case may be, by stimuli of light, darkness or altered temperature. Interrupting continuous darkness by a short period of light, or continuous light by a dark period, or changing from continuous darkness to continuous light, or from continuous light to continuous darkness, or a change in light intensity or even temperature, can initiate rhythmic behaviour. In some cases, a very short exposure to light at some stage in the life-cycle is enough to initiate a rhythm. For example, the fruit-fly, *Drosophila*, normally emerges from the pupa about dawn. If, from the egg stage, the organism is kept in darkness, the young insects emerge at all times of the day. If, however, at any stage between the earliest larval stage and a few days before the emergence of the insect, a brief exposure of light had been given, the insects will all emerge at the time the light stimulus was given. The insect seems to 'remember' the time of the light stimulus as the dawn.

In nature, organisms are exposed to 24-hour periods of alternating light and darkness. Under such conditions the 'rhythmic processes show'

periods of exactly 24 hours. The natural period of the organism appears to get corrected every day to coincide with the solar day. Obviously, this should enable the internal clock to indicate the local time. It is comparable to resetting every morning a watch running fast or slow, to indicate the correct local time. Because of this ability of an organism to adjust its natural period to a 24-hour period, the phase of any rhythm may be caused to bear any desired relationship to the local solar day and night by simply adjusting the periods of light and darkness. For example, if we place an organism under artificial light during night time and in darkness during day-time, the result will be a reversal of the behaviour seen under natural conditions. A cockroach could, thus, be made to run about during day-time and be comparatively quiet during the night. In fact, the internal clock can be shifted by any desired length of time by altering the light-dark periods. Depending upon the organism, the time taken to adjust to the new light-dark cycle in such experiments will vary from 1 or 2 days to more than a week. In human beings, it takes more than a week for a complete phase-shift. This is the reason why it takes some time before

one feels perfectly normal after changing over from a day shift to a night shift or *vice versa*, or while flying from one continent to another. During this interval, the various metabolic processes will get gradually adjusted to the new local time.

In 24-hour light-dark cycles, the natural periods, as already mentioned, will be 'entrained' to exactly 24 hours. Plants and animals are limited in their ability to adapt their rhythms to abnormal light-dark cycles. Organisms can be entrained without difficulty to a 24-hour rhythm and, in some instances, to periods of 22 or 20 hours (for example, in a 11 : 11 or in a 10 : 10 hour cycle of light and darkness). On the contrary, it is well-nigh impossible to entrain most organisms to periods of less than 16 hours or more than 26 hours. Another interesting feature of the ability of a 24-hour light-dark cycle to convert a natural period of about 24 hours to one of 24 hours is that whereas the light period in the cycle may be very short (even a few minutes or seconds), the dark period must be of not less than 3-8 hours. It is not essential that there should be an alternation of light and dark periods for entrainment of rhythms. Cycles of

high and low light intensity will do as well. This is of interest because, under arctic conditions, during summer months, there is no real dark period. It was stated earlier that bees have an awareness of the time at which the flowers they feed upon will secrete nectar and produce pollen. Bees can also be trained to feed at particular times (several times) during the day. But it is impossible to train them to look for nectar at intervals deviating from the 24-hour periodicity, e.g., at intervals of, say 19, 27 or 48 hours.

The periods of endogenous rhythms are not very much affected by changes in temperature. This is surprising since most chemical and physiological processes are affected by temperature fluctuations. It is, however, a very useful attribute to the organism concerned. The clock is the more reliable for being unaffected by temperature changes. In the same way as light-dark cycles of 24 hours are able to alter the natural period of about 24' hours to exactly 24 hours, temperature cycles can also have similar effects. Cycles of high and low temperatures always regulate in such a way that the phase of low temperature coincides with the physiological state usually reached during the night. As in the case

of light-dark cycles, temperature cycles should not deviate too much from the 24-hour periodicity. Otherwise, the physiological rhythm will not follow the temperature rhythm.

The ability of the rhythms to be synchronized by light-dark cycles enables an organism, on being moved from one time zone to another, to have its internal clock reset to the new local time in a few days. From recent studies it has been established that there is a daily rhythm in the sensitivity to changes in light and temperature in organisms. It is this which eventually enables resetting of the clock to the new local time. In its original location the time of higher sensitivity falls during the night. In the new location, since the local light period will overlap the sensitive period in the organism's daily cycle, a slight resetting of the clock occurs due to the higher illumination and temperature at this stage. A similar resetting occurs every day until the sensitive phase again coincides with the night period in the new location. This is believed to be the modus of resetting of the internal clock to the local time.



## Use of the Clock in Navigation

REFERENCE was made earlier to the fact that bees can be trained to feed at specified times during the day. They can also be trained to feed in a different direction at different times of the day. The bees can then remember both the directions and the correct times of the day for each feeding place. This is possible because bees seem to have an instinctive sense of the local time which they are able to determine with reference to the position of the sun. In fact, birds and many other animals like fishes and crustaceans also have the ability to relate the position of the sun to the correct time of the day. This faculty enables them to determine with great accuracy the compass directions and to navigate, using the sun as a compass. The

angle between the sun and any given compass direction gradually changes as the day progresses due to the rotation of the earth. Many organisms employ their solar day clocks to correct their angle relative to the sun, at exactly the rate necessary to compensate for the rotation of the earth.

Many interesting experiments have been performed on bees and birds. The German scientists Martin Lindauer and Karl von Frisch have studied in detail how the bees communicate the information regarding a nectar source to the other bees in a hive. The bee which has located a source makes a wagging dance and describes a figure of '8'. The speed of the dance carries information about the distance of the source from the hive. The shorter the distance, the more rapid the dance. The angle formed by the belly of the bee to the vertical corresponds to the angle formed by the two lines, one of which connects the hive to the sun and the other to the source (*Fig. 7*).

Many birds migrate over very long distances during the change of seasons. For instance, they leave Europe for the southern parts of Africa during autumn and return in spring. These birds follow fixed routes during these migrations.

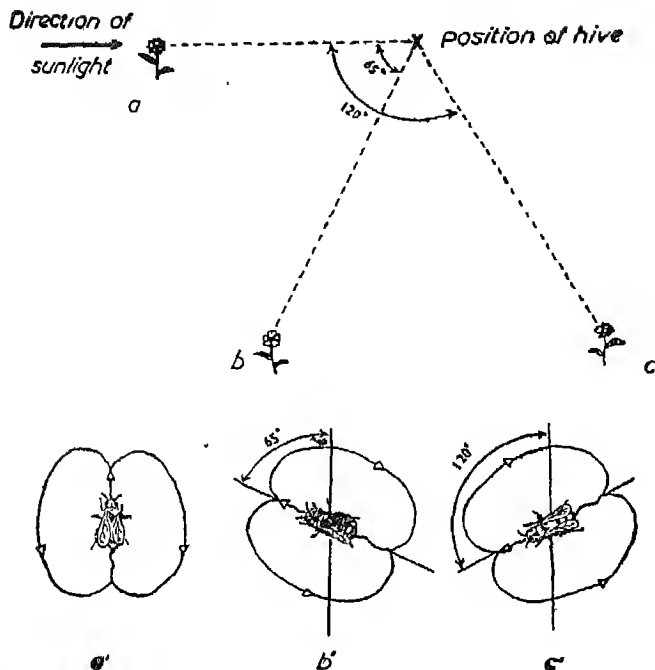


Fig. 7. Method by which a bee indicates the direction of a nectar source. The upper part of the figure shows flowers in three different directions from the hive. The bee does the wagging dance vertically (a) when the flower is in the same direction as the sun; it dances at an angle to the vertical (b) and (c) to indicate that the source lies at that angle with respect to the sun.

Adapted from KARL VON FRISCH,  
 "Dialects in the Language of the Bees",  
*Scientific American*, Aug. 1962, Vol.  
 207, No. 2, 78-87

Experiments under controlled conditions have indicated that the birds are able to do this by using the sun and the stars for compass bearings. That birds can use the sun for finding direction has been shown by experiments in which they were trained to find food in a particular direction and then exposed to a light-dark cycle which was shifted by 6 hours compared to the original day-night cycle. The birds now took off to search for food in a direction deviating from the training direction by  $90^\circ$ . If a bird trained to search for food in a particular direction is held in a domed enclosure having an artificial sun held in a fixed position, it will alter the direction in which it looks for food depending upon the time of the day (*Fig. 8*). Dr Kramer observed in Germany that during the migratory period birds in an aviary in open air became restless and tried to fly off in the migration direction peculiar to the species. This they did only if they could see the position of the sun. They could be made to change this direction if they were deceived by the use of mirrors and given a wrong idea of the direction of the sun (*Fig. 9*). Similarly, birds which migrate at night show a predilection to move in the right direction when placed in a

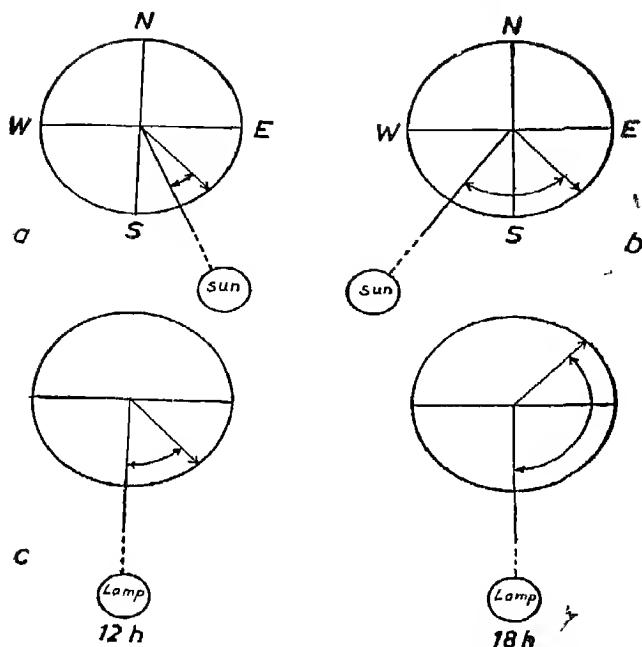


Fig 8. Sun-compass orientation. A bird is searching for food in the SE direction. In (a) and (b) the bird chooses different angles to the sun depending on the time of day. Double arrows show this angle and single arrow the choice of direction. (c) and (d). Corresponding behaviour without sun-light but with a lamp in a fixed position. The bird changes the angle with the static artificial light source during the day. If the direction chosen is SE at 12 h, it is  $90^\circ$  off this direction at 18 h. and consequently, NE.

Adapted from: E. BUNNING, *The Physiological Clock*, Longmans Springer-Verlag, New York, 1967, fig 103

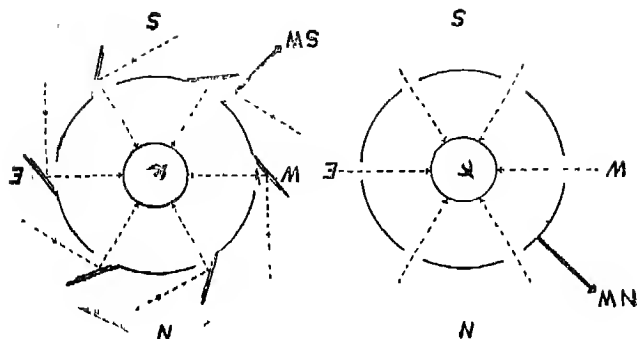


Fig. 9. Kramer's experiments with birds, kept in small cages suspended within a glass-sided enclosure. Left: When sun's rays entered uninterruptedly birds flew in the NW direction. Right: When shutters bearing mirrors were put up so that the sun's rays were deflected through 90°, the birds changed direction through almost 90° and flew in the SW direction.

Adapted from J D CARTHY, *Animal Navigation*, George Allen & Unwin Ltd., London, 1956, fig 51

planetarium only if the star patterns on the ceiling correspond to the local time. By rotating the star patterns the direction of the proposed migration can be altered.

Some ants also seem to be in a position to compensate for the movements of the sun. If they are prevented from continuing their way for

several hours by being placed in a light-tight box, on being released, they move in the original direction showing thereby that they have taken into account the movement of the sun that had occurred during the interval. The wolf spider provides another example for orientation by the sun. This animal lives on the banks of rivers and lakes. When placed in water, it hurries back to the bank in a direction perpendicular to the shoreline. If taken to the opposite bank and placed in water there, it tries to run across the water to the original bank. The direction of movement of this animal is adjusted by the position of the sun. Both ants and wolf spiders can be misled about the position of the sun by means of a mirror as in the case of birds.

Many arthropods probably take note of the incidence pattern of polarized light from the blue sky. Polarized light is light in which all the waves oscillate in one plane. Sunlight reflected from a blue sky is partly polarized, and skylight possesses a distinctive pattern of polarized light which changes with the apparent movement of the sun. At any time of the day, the light coming from the various parts of the sky is characterized by specific amounts and directions of polariza-

tion. Thus the pattern of polarization bears a constant relationship to the sun, and if only a small part of the pattern can be detected, the position of the sun is indicated even when the sun itself may not be seen. The compound eyes of arthropods are particularly well constructed for the reception of polarized light. In fact, red ants have been shown to change the direction of their movement in the laboratory on rotating a polaroid filter placed between them and a source of light above

Sand hoppers living on the wet sands on beaches are believed to be able to orient themselves within reference to the sun as well as the moon.

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## Tidal and Lunar Rhythms

BESIDES the circadian rhythms which are synchronized to the 24-hour day-night cycles under natural conditions, organisms exhibit also other types of rhythms which are regulated by tides and the phases of the moon.

The tides are associated with the period of the revolution of the moon around the earth (29.5 days) and the rotation of the earth relative to the moon (24 hours 50 minutes). The most common tides are semidiurnal. They change direction four times a day, rising from the lowest to the highest and returning again to the lowest twice in 24 hours. Every day, there will be a shift of 50 minutes in the commencement of the flood and ebb tides. The biggest flood tides occur on days when the moon is between the earth and the sun and all the three bodies are in

a line with the forces of attraction of the sun and the moon acting together. The alternation of high and low tides is a special feature of the environment at the edges of oceans. Organisms which inhabit these regions are alternately exposed and covered by the tide. In view of the attendant changes in temperature and light intensity, the organisms inhabiting these zones show tidal rhythms of some sort. Many crabs living in the intertidal zone, for instance, show maximum activity at the time just preceding low tide. Even when removed to the laboratory and placed under constant controlled conditions, the crabs continue to display this rhythm, with the activity synchronizing with the tidal changes at the beach from which they were collected. Similarly, some mussels which pump more water through their gills at the time when the tide is high than during low tide, continue to show the same behaviour in the laboratory under constant conditions so faithfully that one can get an idea of the tidal ebb and flow of their natural habitat by observing these animals.

In addition to the tidal rhythms which are semidiurnal, many organisms also show rhythms of longer duration corresponding to the lunar

cycles. In such rhythms, the maximum and the minimum appear once or twice every lunar month, at the same time. Consequently, these rhythms will have periods of 29.5 or about 15 days. A striking example of monthly rhythm is the behaviour of a small fish called grunion found at the Californian coast. At nights when the forces of attraction of the sun and the moon act together to produce the highest flood tides these fish move towards the sandy beach riding on the crest of the waves. Here, the males and females shed the sperms and eggs in pits that they dig, and get back into the sea with the next wave. The fertilized eggs develop in the moist sand. They are above the water-mark for the next two weeks. During the next spring tide the young fish are released from the eggs and are washed out into the open sea. This goes on with great regularity. *Dictyota*, a brown alga, also shows a lunar rhythm in reproduction, releasing its male and female gametes twice during a lunar cycle, i.e. at intervals of 14-15 days. This periodicity persists even in the laboratory although the alga is not exposed to tidal changes or moonlight. Such cyclic phenomena in reproduction with synchronization of the release of male

and female gametes in large numbers increase severalfold the chances of successful fertilization and the survival of the species. Hence an internal mechanism, timing the activity of organisms to lunar and tidal cycles, is of great advantage.

## Photoperiodism

EXCEPT in the areas of the earth adjoining the equator, there are definite changes in temperature and light conditions corresponding to the change of seasons. Some seasons are markedly cold and others fairly hot. The days are long and nights short during the warmer season and days short and nights long during the cold season. Plants and animals inhabiting these areas are well adapted to these changes in temperature and light conditions. Plants in such areas show a sudden spurt of growth and activity with the onset of spring, culminating in the production of flowers and seeds, by the time the favourable season is at an end. The seeds survive the cold season in a dormant condition. The plants also shed their leaves and cease to grow. The growing points become dormant when the cold season sets in. This dormancy continues till the next spring,

when the growth starts again. This cycle is repeated year after year. The advent of the favourable season leads to mating and reproduction among the animals of these habitats. Such animals tide over the cold season by going into hibernation or, in the case of insects, into diapause.

To be able to survive in these parts from season to season, the organisms, whether plants or animals, should be in a position to anticipate, so to say, the arrival of favourable or unfavourable weather. We have already seen that plants and animals have an innate built-in mechanism to measure out the passage of time during the rotation of the earth around its axis. We now know that the information used by plants and animals to 'predict' the coming season is the alteration in the length of the day that accompanies the change of seasons. The days get shorter as winter approaches and become longer at the beginning of spring. That the length of the day has a profound effect on the growth and development of organisms came to light from observations on certain plants which produced flowers only when the light period during a (24-hour) day was shorter than a certain number of hours. Such

plants are referred to as short-day plants. Other plants, known as long-day plants, flower only when the light period is longer than a certain number of hours. The control of development by the light period is referred to as *photoperiodism*. Photoperiodic effects occur in animals also. The migratory behaviour of birds, growth of wool in sheep, development of sex organs in some animals and the onset or termination of diapause (resting state) in insects are some examples. The beginning of migration of birds can be predicted with an error of only a few days. This makes us believe that birds can distinguish differences in day length by even a few minutes. Plants are also able to measure day length as accurately. For example, in the case of the rice plant, it has been observed that a difference in day length of only one minute results in an acceleration or inhibition of development by more than one day!

The photoperiodic effect has been shown to be independent of temperature to a large extent. This is as it should be, since the day length on any particular day at any particular place is a constant factor, but not the temperature, and if the innate mechanism measuring day length is

to be reliable, it should not be affected by changes in temperature. Again, if the photoperiodic information about seasonal changes is to be dependable, the intensity of light should not have much effect. This is actually the case, and plants and animals actually measure time from the moment the sun is still a few degrees below the horizon in the morning to the moment when it is again a few degrees below the horizon after sunset. However, the intensity of moonlight is not high enough to bring about an error regarding the length of the photoperiod.

In its accuracy and temperature-insensitivity the mechanism of day length measurement resembles the endogenous clock measuring the passage of time during a 24-hour day. That the two mechanisms are basically the same has been demonstrated from experiments which have revealed that sensitivity to light in certain photoperiodic processes fluctuates with a 24-hour periodicity. For example, when a long-day plant was grown with days 9 hours and nights 39 hours long, light interruptions early in the dark period promoted flowering; the effectiveness of the interruptions increased to a maximum as the interruption was placed later



and later ; then the effect started decreasing. Still later, interruptions again increased flower-

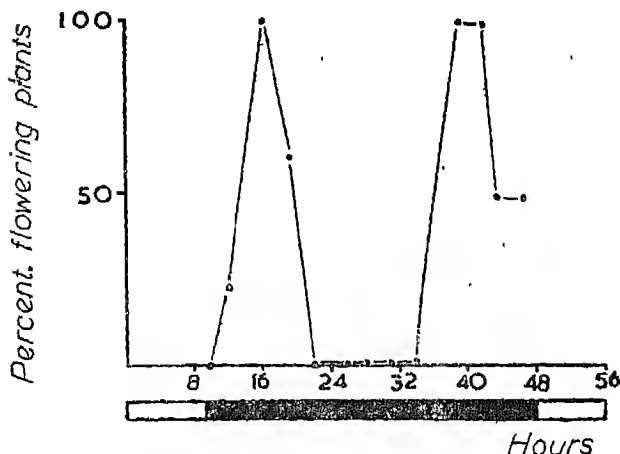


Fig 10. Effect of light breaks of two hours at period on flowering in a long-day plant. Times of maximum promotion of flowering are seen to occur 24 hours apart. The horizontal strip below indicates 9 : 39 hour light-dark cycles

Adapted from E BÜNNING, *The Physiological Clock*, Longmans 'Springer-Verlag, New York, 1967, fig. 121

ing to a second maximum. The two peaks of maximum flowering were found to be 24 hours apart (*Fig. 10*). Similar results have been obtained with insects when the diapause-inhibit-

ing effect of light interruptions during the dark phase was studied. Thus, there appears to be a cyclic fluctuation in sensitivity to light in the photoperiodic phenomena. This is comparable to the situation in diurnal rhythms (see p. 24). In *Kalanchoe*, a short-day plant, the inhibiting effect (in initiation of flowering) of light interruption during the dark period is maximum at the time when flowers, if present, show maximum closure. Apparently, it is the same timing mechanism that is responsible for photoperiodic effects and diurnal rhythms.

Organisms have to measure not only the day length but also the direction in which the day length changes, i.e. whether it is increasing or decreasing. This is essential because, except for the shortest day and the longest day, each day-length occurs twice a year, once between winter and summer, and a second time between summer and the next winter. There is probably a built-in necessity for short days or long days to precede long days and short days, respectively, for the appropriate photoperiodic effect.

## Mechanism of the Clock

IN CONSIDERING the mechanism of the biological clock it is important to know at first whether the diurnal rhythms seen in plants and animals are truly endogenous or only exogenous. As already mentioned, all organisms are, under natural conditions, exposed to diurnal fluctuations of light and temperature. However, the persistence of the rhythms, at least for some time, under constant conditions of light (or darkness) and temperature with characteristic periodicities of *about* 24 hours rules out the possibility that diurnal rhythms are due to the fluctuations in external conditions. But some scientists are inclined to explain the persistence of the rhythms under controlled conditions as due to the effect of other subtle and pervasive geophysical forces such as atmospheric pressure, cosmic radiation

and the earth's magnetic field, which cannot be controlled easily in the laboratory.

That the clock is present even at the cellular level is clear from the fact that many unicellular plants and animals show various types of rhythmic phenomena. Isolated plant parts like leaves and tissue cultures also exhibit rhythms.

There have been speculations connecting rhythmic phenomena with biochemical activities in cells. Circadian metabolic rhythms reflect rhythms in enzyme activity. For instance, the enzyme luciferase, responsible for the luminescence in the alga *Gonyaulax*, fluctuates diurnally. However, such fluctuations probably do not represent the actual clock mechanism. They are only processes controlled by the clock just like changes in growth rate, turgor pressure or running activity. The length of the periods or their parts is related to the intensity of metabolic activity. Enzyme inhibitors can decrease the amplitude of the rhythms but do not suppress the clock. For instance, sodium cyanide reduces the number of spores discharged by the alga *Oedogonium*, but when the concentration of the poison decreases gradually spore discharge increases again. The periodicity in spore release is not affected by the

poison. Substrates or an increase in temperature can accelerate the rate of metabolic processes but do not, as we have already seen, affect the rhythms appreciably.

Attempts have also been made to identify the clock with particular organelles in the cell. In view of its vital role in many of the activities of the living cell, the nucleus has been an attractive candidate among the various cell organelles. Diurnal fluctuations in the volume of the nucleus has been observed in both plant and animal cells. However, experiments with the marine alga *Acetabularia* have shown that certain diurnal rhythms like those in photosynthesis continue for several days under constant conditions in cells from which the nucleus has been removed. The nucleus has, nonetheless, an important role in circadian rhythms in *Acetabularia*, as shown by the fact that when two plants exposed to opposite light-dark cycles for more than 14 days have their upper parts interchanged, the rhythm of oxygen evolution seems to be determined by the nucleus at the base of the plant and not by the upper part.

It is the nucleus which regulates the synthesis of enzymes and other proteins in cells. This

is achieved by the formation of a substance called ribonucleic acid (RNA) which is released from the nucleus. This RNA, called messenger RNA, is formed on the basis of information carried in the genetic material of the nucleus in the shape of deoxyribonucleic acid (DNA). Based on the information carried by the messenger RNA, an appropriate protein is formed in the cytoplasm by the cooperation of a series of enzymes and organelles. The formation of messenger RNA can be inhibited specifically by certain poisons. These poisons also suppress some endogenous rhythms suggesting the probability that the clock function is dependent upon RNA synthesis. Only future investigations can unravel the true nature of the clock mechanism in living cells.

In animals, secretions from the nervous system called neurohormones and from endocrine glands seem to have a controlling effect on some of the rhythmic phenomena. The activity rhythm in the cockroach seems to be controlled by secretions from the suboesophageal ganglion (a mass of nerve cells in the head region). Removal of the head of the cockroach results in loss of activity rhythm. If the suboesophageal ganglion from a normally rhythmic cockroach is trans-

planted into a headless cockroach, the activity rhythm is restored. It is the brain which responds to photoperiodic stimulus and controls development in insects as shown by experiments on some aphids. In the normal insect, light falling on the head alone can evoke photoperiodic response. If the brain is excised and reimplanted in the tip of the abdomen, the posterior of the insect acquires that ability. Endocrine glands like the adrenals and the pituitary show rhythmic activity. The diurnal fluctuations in eosinophil and leucocyte counts in blood are controlled by the hormones secreted by the adrenals. The effect of adrenal secretions on asthmatic attacks was referred to earlier.

In the higher organisms, the role of the endocrine glands and the central nervous system seems to be to exert a controlling effect on the endodiurnal oscillations of the individual organs and tissues and the synchronization of the processes in different parts of the body. Such synchronization does not necessarily result in maxima and minima occurring at the same times in the different parts or organs. But the different rhythms bear a certain relationship to one another. This is very essential for the

healthy functioning of the body as demonstrated by some very interesting experiments with the cockroach. In this insect, when the suboesophageal ganglion from one individual is implanted into another, tumours develop in the midgut if the two individuals had been exposed to opposite light-dark cycles, i.e. if the two cockroaches had rhythms out of phase with each other. When a person travels by air to a new time zone, all the rhythms in the body do not adjust themselves to the new light-dark cycle at the same time, some doing so earlier than the others. It is for this reason that one takes a few days to feel quite normal after such travel

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## Practical Applications

THE STUDY of biological rhythms has led to some practical applications in the fields of agriculture and medicine. A knowledge of the light requirements for flowering enables the controlling of the light-dark regimes suitably in greenhouses so that an annual plant could be made to bear fruits all round the year. Similarly, by providing additional light artificially in the field long-day plants could be made to flower even under short-day conditions. In greenhouse cultures of plants, addition of nutrients may be made to coincide with the time of their optimum uptake by roots. Feeding rhythms for fish in small aquaria have been studied and the information utilized in providing food at the proper times. The fact that short days increase the growth of wool in sheep and fur in fur-yielding animals has been used to good advantage. The

sheep are put in a dark shed during part of the day during summer months to increase the yield of wool. In poultry, increased egg production has been achieved by artificially lengthening the day (with electric lights).

The diurnal changes in the composition of the blood makes it necessary to choose the appropriate time for treatment of certain ailments with hormones like adrenalin and insulin. It is dangerous to inject adrenalin into a person when blood level of the hormone is at a maximum and sugar level at a minimum. In diabetes, insulin should be administered when sugar level in the blood is the highest. The diurnal changes in cortecosteroids in the blood and their effects on asthmatic attacks have already been referred to. The administration of cortisone or similar drugs should be timed to give the most beneficial results. This holds good also in the treatment of hypotension or hypertension. It is to be remembered that blood pressure is lowest in the morning and highest in the evening.

A study of the physiological rhythms in human beings has become even more important now when man has actually started travelling out into outer space where the diurnal changes in the

environment that we experience on the earth are totally absent. The concept of time based on the 24-hour day will not be valid in outer space. How the human system behaves outside the synchronizing effect of the 24-hour day-night cycle of the earth is of great significance, particularly when travel into outer space extending over long periods is planned.

## References and Suggested Reading

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